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- 109-132** **PREBIOTIC MECHANISMS, FUNCTIONS AND APPLICATIONS – A REVIEW**
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ABSTRACT: *In October 2012, a group of scientists met at the 10th Meeting of the International Scientific Association of Probiotics and Prebiotics (ISAPP) in Cork, Ireland to discuss issues surrounding prebiotics and their development. This article summarises outputs from the meeting. Various prebiotic definitions were discussed and how the concept has evolved from targeting the colonic microbiome, through to the entire gastrointestinal tract and finally the ISAPP definition, which specifies fermentation as a key criterion. Structure and function relationships are becoming clearer with effects upon microbial diversity, determinations of selectivity and enhanced biological activity being major outcomes. Immune modulation and metal chelation were further facets. Biomass can be a useful, and economic, means of generating new prebiotics. Pectic oligomers from citrus are model examples. Testing aspects range from in vitro batch culture fermenters to multiple stage models, immobilized systems, animal, cellular studies and human trials. Analytical processes around microbiota characterization and functionality were compared. Human studies were seen as the definitive outcome, including ¹³C labeling of key interventions. For extra intestinal effects, atopic disease, respiratory infections, vaginal issues, oral disease, adiposity, liver damage and skin infections are all feasible. The general outcome was that microbiota modulation was the key mechanism that linked these interactions. In pet food applications, the market potential for prebiotics is huge. Health targets are similar to those of humans. Issues include monomeric composition, chain length, linkages, branching, microbiota beyond bifidobacteria, metabolic function, mechanisms of health effects. Molecular biology has unraveled some of the explanations for prebiotic influences e.g. gene clusters to show transporters, regulators, permeases, hydrolases, lacS. In *Lactobacillus ruminis*, fermentation studies have been aligned to genome annotations, showing an energy efficient and rapid transport of GOS. In bifidobacteria, functional genome analyses have demonstrated uptake of trisaccharides. Questions relating to patients were then raised. For example, are prebiotics related to disease treatment or health maintenance? If a prebiotic does not change the microbiota, then how does it operate? Case study trials in Inflammatory Bowel Disease were presented on patient access to prebiotics and information. These showed that their knowledge of prebiotics was poor, compared to probiotics. The group then discussed the next generation of prebiotics (e.g. anti-adhesive activities). The comparator was Human Milk Oligosaccharides, which both reduce adherence of pathogens and act as prebiotics. Studies with galactooligosaccharides (GOS) have used pyrosequencing to demonstrate varying species level effects. This has relevance for infant formulae. Prebiotic aspects of whole foods and their complexity was covered. Trials were described where cross feeding and co-metabolism had been investigated. Suggestions on other prebiotic influences, aside from bifidobacteria, were made and included metagenomics, metabonomics, gene expression, mRNA global sequencing, bile deconjugation, enzyme profiles, lipids, phenolics. The discussion suggested how prebiotics could move forward with a wider expansion of the concept, target populations, expanded microorganisms, health benefits, application of new technologies and improved consumer understanding being the main goals.*

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- 133-144** **AN ASSESSMENT OF THE PREBIOTIC POTENTIAL OF SINGLE AND BLENDED SUBSTRATES IN ANAEROBIC *IN VITRO* BATCH CULTURE FERMENTATIONS USING CANINE FAECAL SAMPLES AS INOCULA**
V. L. Inness, C. Khoo, K. L. Gross, L. Hoyles, G. R. Gibson and A. L. McCartney

ABSTRACT: *Previously, using in vitro static batch culture, it was found that rice bran (RB), inulin, fibersol, mannanoligosaccharides (MOS), larch arabinogalactan and citrus pectin elicited prebiotic effects (in terms of increased numbers of bifidobacteria and lactic acid bacteria) on the faecal microbiota of a dog. The aim of the present study was to confirm the prebiotic potential of each individual substrate using multiple faecal donors, as well as assessing the prebiotic potential of 15 substrate blends made from them. Anaerobic static and stirred, pH-controlled batch culture systems inoculated with faecal samples from healthy dogs were used for this purpose. Fluorescence in situ hybridization (FISH) analysis using seven oligonucleotide probes targeting selected bacterial groups and DAPI (total bacteria) was used to monitor bacterial populations during fermentation runs. High-performance liquid chromatography was used to measure butyrate produced as a result of bacterial fermentation of the substrates. RB and a MOS/RB blend (1:1, w/w) were shown to elicit prebiotic and butyrogenic effects on the canine microbiota in static batch culture fermentations. Further testing of these substrates in stirred, pH-controlled batch culture fermentation systems confirmed the prebiotic and butyrogenic effects of MOS/RB, with no enhancement of Clostridium clusters I and II and Escherichia coli populations.*

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- 145-148** **PREVENTION OF RELAPSE FOLLOWING *CLOSTRIDIUM DIFFICILE* INFECTION USING PROBIOTIC *LACTOBACILLUS CASEI* SHIROTA**
Lennard YW Lee, Ramez Golmohamad, and George MacFaul

ABSTRACT: *Once patients have had Clostridium difficile infection (CDI), recurrence rates are high with many patients experiencing a relapse of their disease. Lactobacillus casei Shirota is a probiotic that reduces rates of antibiotic-associated diarrhoea. There have been no studies analyzing the use of this probiotic in patients who have had an episode of CDI to prevent relapse. This study was a single site, cohort-control study of patients with CDI and treated with either antibiotics alone or antibiotics and probiotics (L. casei Shirota). 66 patients were included for analysis in this study, 31 had probiotics and antibiotics and 35 who had no-probiotics. The median age of the patients was 78 years and 33.3 % were male. Rates of recurrent CDI were significantly lower in the probiotics cohort, 3.2% vs 20.0% (p=0.007). We conclude, patients who have had CDI have a high rate of early re-admissions to hospital with significant morbidity and mortality. This study suggests that the use of the widely available probiotic strain L. casei Shirota at the time of initial infection may be associated with lower rates of CDI recurrence and lower readmission rates. Further prospective studies are required.*

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149-156 GASTROINTESTINAL SURVIVAL OF BACTERIA IN COMMERCIAL PROBIOTIC PRODUCTS

Mathieu Millette, Anne Nguyen, Khalie Mahamad Amine and Monique Lacroix

ABSTRACT: *This work compared bacterial gastrointestinal (GI) resistance of commercial probiotic products (capsules, fermented milk and powder). To simulate GI transit, the probiotic products were subjected to gastric fluid for 120 min then to intestinal fluid for 180 min. Gastric and intestinal fluids were prepared according to United States Pharmacopeia protocols. Bacterial enumeration was compared before and after the GI transit to evaluate the protective effect of the vehicle or the food matrix. Bacteria of the four probiotic capsules covered with an enteric coating had a higher survival rate (<1 log₁₀ CFU reduction) than uncoated. Eight encapsulated but non enteric coated probiotic products showed limited GI resistance (between 1 and 5 log₁₀ CFU reduction) while five products showed no GI survival. For probiotic fermented milk, two products demonstrated excellent or good protective property (<1 log₁₀ CFU reduction) while the other four showed no resistance. Only one of six powdered probiotic strains had excellent GI survival. This study demonstrated that GI survival varies from one probiotic product to another. It reiterates the importance of manufacturing probiotic strains using the appropriate vehicle for the bacteria to reach its site of action and produce the expected beneficial effects.*

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157-164 CHARACTERIZATION OF PROBIOTIC LACTIC ACID BACTERIA TO BE USED AS STARTER AND PROTECTIVE CULTURES FOR DAIRY FERMENTATIONS

Gamal Enan, Seham Abdel-Shafi, M. F. Abdel-Haliem and S. Negm

ABSTRACT: *The study employed herein was an endeavor to select probiotic starter and protective cultures of lactic acid bacteria. One hundred strains of lactic acid bacteria isolated from Zabady samples (Arabian yoghurt) were surveyed by common methods for growth, acidification of medium and inhibition of some food-borne pathogens. Three isolates only named: Z11, Z20, Z55 showed the desired properties. Based on their phenotypic and biochemical properties as well as sequences of 16 SrRNA gene of each strain, Z11 strain was identified as belonging to *Lactococcus Lactis* subspecies *Lactis* (*L. lactis* Z11) and both Z20 and Z55 were identified as belonging to *Lactobacillus delbreukii* subspecies *bulgaricus* (*Lb. bulgaricus* Z55). Maximal values of growth (CFU/ml) and medium acidification (final pH) of *L. Lactis* Z11 and *Lb. bulgaricus* Z55 were obtained in broth cultures adjusted at 30 oC ; 42 oC and final pH values were 4.3; 3.8 respectively after 24 hours of incubation. Both *L. Lactis* Z11 and *Lb. bulgaricus* Z55 showed promised positive results with regard to lactose metabolism, proteolytic activity, Voges Proskauer test, production of acetoin, citrate utilization, β and δ glycosidase activity, production of D (-) and L (+) isomers of lactic acid, tolerance to 4% NaCl, coagulation of milk, glutamate decarboxylase and inhibition of some food-borne pathogens.*